

## Rhinocerebral zygomycosis in a sheep

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**A** 2-year-old, Barbados black belly sheep was admitted to the Veterinary Teaching Hospital, School of Veterinary Medicine, The University of the West Indies, Trinidad, with a history of sudden onset of anorexia, extreme dyspnea, and unilateral mucohemorrhagic nasal discharge.

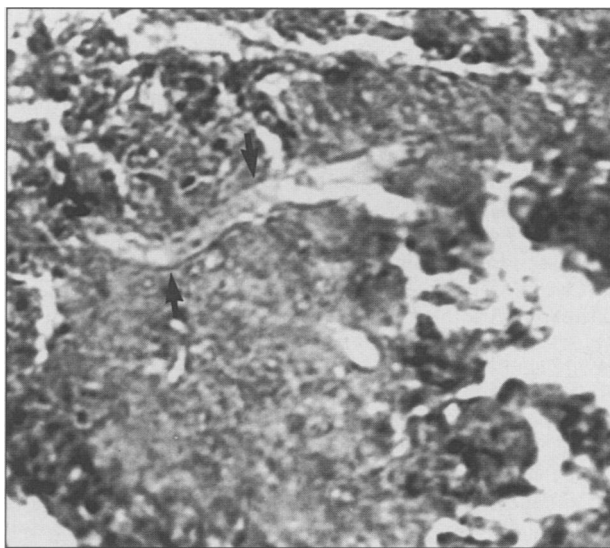
The animal came from a flock of 60 sheep kept for weed control around a homestead on approximately 10 000 m<sup>2</sup> (2.5 acres) of land. A ravine runs through the land, but the owner reported that it is not prone to flooding. During daytime, animals are kept in pasture or open yard, and at night, they are kept in wooden crates in a covered shed. The animals are fed grass ad libitum (a combination of pasture and zero grazing), supplemented with byproduct-based concentrate ration. Recently, 3 rams from the same farm, 1 to 3 y of age, had been presented to the local veterinarian with signs varying in severity, including fairly sudden onset of respiratory signs characterized by mucohemorrhagic nasal discharge and facial pain on percussion of frontal and nasal sinuses. Marked respiratory distress; locomotory incoordination; and blindness, in the most severe case, were observed. The animals were depressed and had poor appetite, with marked difficulty in prehension, mastication, and swallowing. Sometimes, impacted ingesta had to be removed manually from the oral cavity and oropharynx. Facial distortion was not noted in any of these cases. The animals were treated with trimethoprim-sulfamethoxazole (Trisal-kel 240; Kela Laboratories, Hoogstraten, Belgium), 44 mg/kg body weight (BW), IM, but the condition did not improve and 2 of the 3 animals eventually died.

The animal referred to the Veterinary Teaching Hospital was in poor body condition and had unilateral mucohemorrhagic nasal discharge. It appeared blind and deaf, had ruminal impaction, and kept its hind legs tucked forward under the body in an unsteady stance, suggesting locomotory incoordination. Additional clinical manifestations during hospitalization included head pressing and teeth grinding. Radiographs revealed a round radiopacity, approximately 3 cm in diameter, in the region of the dorsal and lateral conchae, bordered caudally by the cribriform plate of the ethmoturbinate bones. An increased radiopacity of the frontal sinus was also observed.

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**Figure 1.** Periodic acid-Shiff stained section of necrotic areas from the ethmoturbinate bones, showing fungal hyphae (arrows). Magnification 400X.



**Figure 2.** Lactophenol cotton blue staining of the *Conidiobolus* sp. isolate, showing characteristic thick-walled zygosporangia formed by the union of 2 contiguous hyphal cells (left). A discharged sporangiole with a basal papilla and a hyphal cell emerging from it was observed (right); bar = 10 µm.

A sample of the nasal discharge was submitted for bacteriological culture, and the animal was placed on a combination of trimethoprim-sulfamethoxazole, 44 mg/kg BW, and dexamethasone (Dixazone; Rhône-Mérieux, Victoriaville, Quebec), 6 mg, administered IM. The bacteriological culture yielded *Mannheimia haemolytica* (formerly *Pasteurella haemolytica*) along with other *Pasteurella* spp. However, the animal's condition was deteriorating so rapidly that it

was euthanized with an IV injection of sodium pentobarbital, 2 d after admission.

At postmortem, the dorsal and ventral conchae were hyperemic and the meatuses were filled with fibrinous material. The ethmoturbinate bones were necrotic. A perforation of about 1 cm in diameter through the cribriform plate of the ethmoid bone connected the right nasal cavity with the cranial cavity. The frontal sinus was filled with fibrinous material similar to that found in the meatuses. The brain appeared grossly normal, except for a portion of the frontal lobe that was necrotic.

The rumen was impacted and there was erosion of the mucous membrane of the pyloric sphincter, approximately 3 cm in diameter. Other vital organs, such as the lungs, liver, spleen, pancreas, kidneys, and gastrointestinal tract, appeared normal.

On microscopic examination, the frontal cortex of the brain was necrotic with a marked infiltration of inflammatory, predominantly mononuclear, cells. There was diffuse necrosis with extensive infiltration of neutrophils and mononuclear cells, including macrophages and plasma cells. Numerous micro-abscesses were evident and multinucleated giant cells were also present. Fungal hyphae were seen in periodic acid-Schiff (PAS)-stained sections of diffuse necrotic areas of the ethmoturbinates and the frontal cortex (Figure 1). Histological sections from lungs, liver, spleen, pancreas, kidneys, and gastrointestinal tract were normal.

Different tissues, including the fibrinous material from the meatuses, were inoculated in duplicate onto 5% sheep blood, MacConkey's agar, and Sabouraud's dextrose agar. Plates were incubated at room temperature (approximately 25°C) and at 37°C. Fungal growth was observed on both blood and Sabouraud's dextrose agar plates after 48 h of incubation at room temperature.

The colonies grew rapidly on both blood and Sabouraud's dextrose agar. On the latter medium, colonies had a circular thallus reaching a diameter of approximately 6 cm after 1 wk. The colonies, which were initially flat and membranous, became irregularly folded and wrinkled with age. They were pale and yellowish-white. Production of aerial mycelium was scanty. On sheep blood agar, the colonies produced a wide zone of beta hemolysis and appeared

grey to khaki, flat, glabrous with radial folding. On lactophenol cotton blue staining, the organism produced conidia that appeared round with a papillary protuberance characteristic of a *Conidiobolus* sp. In addition, numerous spherical, thick-walled zygospores were observed, as well as hyphal tubes germinating into conidiophores (Figure 2).

*Conidiobolus* spp. (class Zygomycetes, order Entomophthorales) are generally found in the environment, mostly on decaying vegetation. Although infections with various *Conidiobolus* spp. have been reported, *C. incongruus* and *C. lamprauges* have been associated, to some extent, with cases of mycotic infections in different animal species, including humans, in subtropical and temperate regions (1–5).

Based on these reports, diseases caused by zygomycetes, in general, and *Conidiobolus incongruus*, in particular, appear frequent in Australia, where 2 outbreaks have been reported in sheep (3,5) and, more recently, a case in a deer (4). With few exceptions, only the upper respiratory tract and surrounding tissues have been involved.

Our case resembles those reported from Australian outbreaks (3,5). However, although the characteristics of the infecting agent on blood agar was described in one of these reports (5), production of hemolysin was not mentioned.

## Acknowledgment

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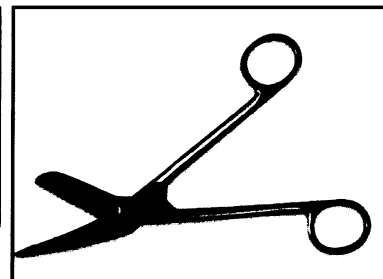
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